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## Procedures for the successful regeneration of leafy spurge

DAVID G. DAVIS

USDA-ARS, Metabolism and Radiation Research Laboratory, State University Station, Fargo, ND 58105.

Tissue culture systems were used to study the process of organ formation in leafy spurge. The intent is to try to standardize conditions that will result in plant formation from individual cells or cell clumps, and then to apply an environmental or chemical stress on the system. The cells are monitored for the formation of roots and/or shoots. If physiological processes involved in organ formation are found that are unique to leafy spurge, and if they can be interfered with by chemical or biological methods, the control of the weed might be accomplished.

Plants have been regenerated from cell suspensions in one of seven accessions of leafy spurge. Plants have been transferred from the tissue cultures to the greenhouse, and the plants look similar to the parent plant. Externally applied plant growth regulators altered growth patterns, but not in any consistent way. The cell suspensions grow actively in the presence of 2,4-D. Regeneration occurs if the 2,4-D is removed and the cells are washed free of old media and maintained in hormone-free media. The responses in various experiments differ dramatically, and to date no medium or growth condition has proved consistent with reliable and reproducible results, but several specific conditions appear to work most of the time. White light (cool white fluorescent) generally enhances root and shoot formation, and filtered light (transmission maxima of 450 or 650 nm) often increases root formation. From 50 to 67% of the inorganic nitrogen should be supplied as nitrate and about 13-millimolar potassium usually work well. The total nitrogen can vary considerably. Eighteen to 27 mM works well, although 60 mM can also be used.

Regenerated plants growing on agar or in liquid in flasks contain some epicuticular wax platelets. Cells from both organized and unorganized tissues contain large amounts of densely stained material within the vacuoles. Large numbers of microbodies with well-formed crystals are present in young plantlets developed from liquid cultures.